COMPOSITIONS AND METHODS FOR REDUCING BACTERIAL AGGREGATION

RELATED APPLICATIONS PARAGRAPH

[0001] This application is a continuation of International Patent Application No. PCT/AU2019/050893, filed Aug. 23, 2019, entitled "Compositions and Methods for Reducing Bacterial Aggregation". Foreign priority benefits are claimed under 35 U.S.C. § 119(a)-(d) or 35 U.S.C. § 365(b) of Australian Application No. 2018903096, filed Aug. 23, 2018. The contents of each of these applications are incorporated herein by reference in their entirety.

FIELD OF THE DISCLOSURE

[0002] The present disclosure relates to compositions and methods for inhibiting bacterial aggregation and in particular to compositions and methods that inhibit autotransporter-mediated bacterial aggregation or attachment.

BACKGROUND OF THE DISCLOSURE

[0003] Any discussion of the prior art throughout the specification should in no way be considered as an admission that such prior art is widely known or forms part of the common general knowledge in the field.

[0004] Biofilms are complex communities of bacteria living in close association with each other and a surface. Compared to planktonic cells, bacteria which are protected within a biofilm display resistance to conventional antibiotics, biocides and hydrodynamic shear forces (Bjarnsholt et al., Nat. Rev. Drug Discov. 2013. 12: 791-806).

[0005] Biofilms are significant threats in medical, industrial and environmental settings. Biofilms in the environment can lead to the persistence of foodborne pathogens. For example, biofilm formation by enterohemorrhagic *E. coli* (EHEC) O157:H7 can occur on plant surfaces (Torres et al., Appl. Environ. Microbiol. 2005. 71: 8008-15; Choi et al., J. Appl. Microbiol. 2011. 111: 1465-72), and more than 25% of outbreaks caused by these zoonotic shiga toxin-producing pathogens originate from contamination of commercial produce such as lettuce, spinach, cabbage, sprouts or tomatoes (Rangel et al., Emerg. Infect. Dis. 2005. 11(5): 603-9). In industrial settings, EHEC biofilm formation has also been observed on abiotic surfaces such as stainless steel, glass and plastic (Torres et al., Appl. Environ. Microbiol. 2005. 71: 8008-15; Dourou et al., Int. J. Food Microbiol. 2011. 149: 262-8).

[0006] Many bacterial infections in humans are associated with bacterial aggregation and biofilms. Respiratory and urinary tract infections, infections on medical devices and infections of the ear, gums and heart have all been associated with bacterial biofilms. Uropathogenic *E. coli*, for example, are responsible for 75 to 95% of all uncomplicated urinary tract infection (UTI) cases (Hooton, N. Engl. J. Med. 2012. 366(11): 1028-37). These infections cause significant morbidity and are of increasing concern due to the emergence of multi-drug-resistant strains (Totsika et al. 2012. Curr. Drug Targets 13(11): 1386-99).

[0007] Biofilms act to shield bacteria from host immune factors, as well as from antibiotic agents such as antimicrobial drugs and chemical detergents. Infections caused by bacteria that grow as aggregates in biofilms are therefore often chronic as they resist innate and adaptive defence mechanisms as well as antibiotics. Moreover, it has been suggested that as the aggregated bacteria in chronic infections are in close proximity to one another, genes coding for resistance to antibiotic agents can be passed horizontally from one bacterium to another (Bjarnsholt et al., Nat. Rev. Drug Discov. 2013. 12: 791-806). Current treatments for biofilm-associated infections include surgical removal of infected tissue or medical indwelling. Antibiotic agents are also used, however, they are often ineffective due to the shielding effect of the biofilm and due to the reduced metabolic activity of the aggregated bacteria.

[0008] In this context, there is a need for compositions and methods for reducing bacterial aggregation or biofilm formation.

SUMMARY OF THE DISCLOSURE

[0009] In work leading to the present disclosure, the inventors observed that a class of outer membrane and secreted proteins called autotransporters contribute to bacterial aggregation, biofilm formation and bacterial attachment to surfaces. Using structural, biochemical and functional techniques, the inventors found that homodimerisation of bacterial autotransporter proteins enables bacteria to aggregate and form biofilms. The inventors also found that autotransporter proteins contribute to bacterial attachment to surfaces. As described herein, the inventors have developed autotransporter-binding molecules which block autotransporter interactions and inhibit bacterial aggregation and biofilm formation.

[0010] In a first aspect, the present disclosure provides an isolated antibody or antigen binding fragment thereof comprising:

[0011] a) a CDRH3 comprising the sequence set forth in SEQ ID NO: 5 or a CDRL3 comprising the sequence set forth in SEQ ID NO: 8; or

[0012] b) a CDRH3 comprising the sequence set forth in SEQ ID NO: 17 or a CDRL3 comprising the sequence set forth in SEQ ID NO: 20.

[0013] The isolated antibody or antigen binding fragment may comprise:

[0014] a) a CDRH3 comprising the sequence set forth in SEQ ID NO: 5 and a CDRL3 comprising the sequence set forth in SEQ ID NO: 8; or

[0015] b) a CDRH3 comprising the sequence set forth in SEQ ID NO: 17 and a CDRL3 comprising the sequence set forth in SEQ ID NO: 20.

[0016] The isolated antibody or antigen binding fragment may comprise:

[0017] a) a CDRH1 comprising the sequence set forth in SEQ ID NO: 3;

[0018] a CDRH2 comprising the sequence set forth in SEQ ID NO: 4;

[0019] a CDRH3 comprising the sequence set forth in SEQ ID NO: 5;

[0020] a CDRL1 comprising the sequence set forth in SEQ ID NO: 6;

[0021] a CDRL2 comprising the sequence set forth in SEQ ID NO: 7; and

[0022] a CDRL3 comprising the sequence set forth in SEQ ID NO: 8; or

[0023] b) a CDRH1 comprising the sequence set forth in SEQ ID NO: 15;

[0024] a CDRH2 comprising the sequence set forth in SEQ ID NO: 16;